In the name of Allah, the Most Gracious, the Most Merciful

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Outils du génie génétique :

- **Rôle** : manipulation d'un gène

I) Les outils

1- Enzymes de restriction :

- **Rôle** : couper l'ADN

endo ↓

3' ATCG TA 5'
5' TAGC AT 3'
(T 3' entouré : exo)

a) Endonucléases :

Reconnaissent : un site de restriction de nature Palindromique.

5' ATGCGCAT 3'
3' TACGCGTA 5'

↓

5' AT 3' 5' GCGCAT 3'
3' TACGCG 5' TA 5'

Extrémités cohésives

ATGC GCAT
ATCG CG TA → 5' ATGC 3' 5' GCAT
3' TACG 5' 3' CGTA

extrémités Franches

b) Exonucléases :

5' ATCGAT 3'
3' TAGC (T)(A) 5'
↙ ↙
-T -A

2- Ligases :

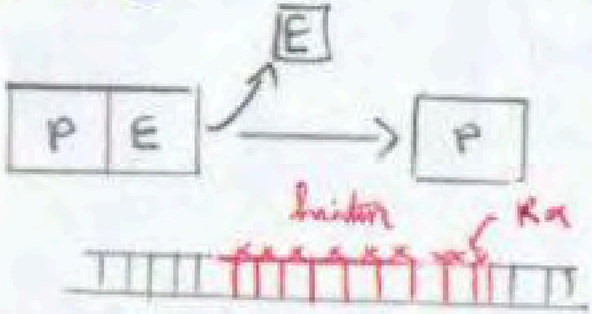
lient les segments d'ADN par des liaisons phosphodiester

3- ADN polymérases :

Σse de l'ADN

a) fragment de Klenow :

ADN polymérase I sans activité exonucléasique 5' → 3' à 37 C



Pour le marquage

b) Taq Polymérase :

70 C° : Technique PCR (Amplificat° de l'ADN)

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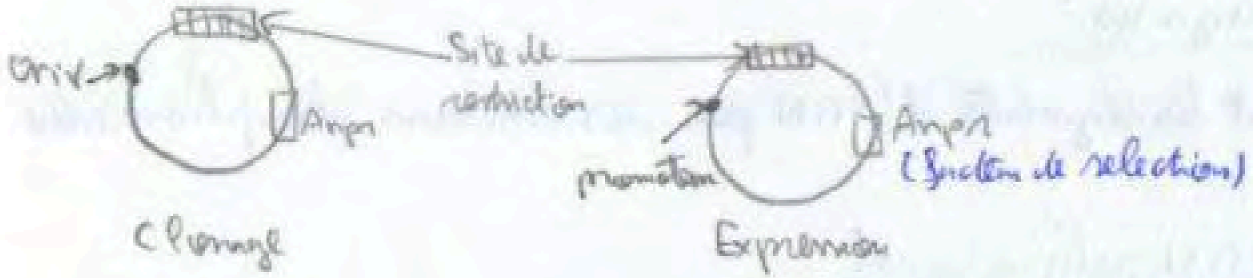
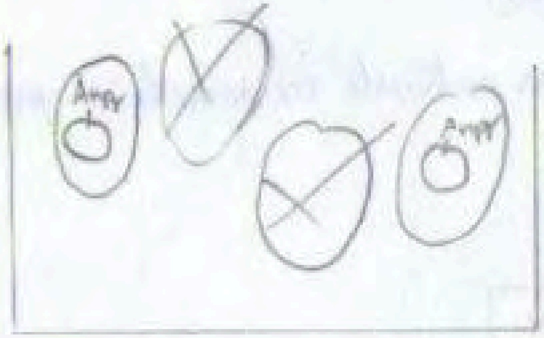
4) Vecteur:

Transporteurs de gènes.

- $\vec{V}$ de clonage : clone un gène
- $\vec{V}$ d'expression : expression d'un gène



Ampr : gène de restriction à l'Ampicilline



5) Sonde moléculaire

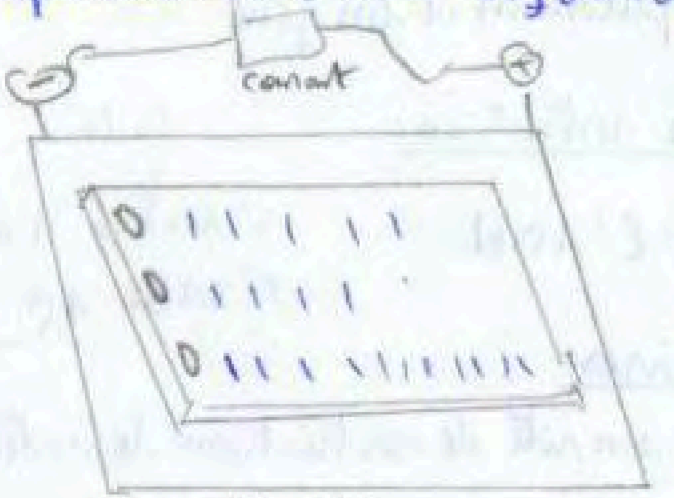
Séquence d'ADN monocaténaire marquée, complémentaire du gène recherché

CTAGC* : Sonde

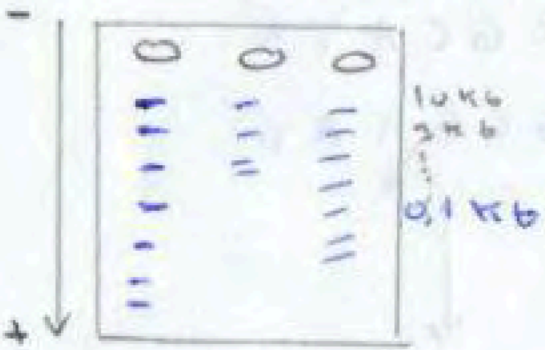
3' ATCCGATC GAACT 5'

6) Technique d'électrophorèse:

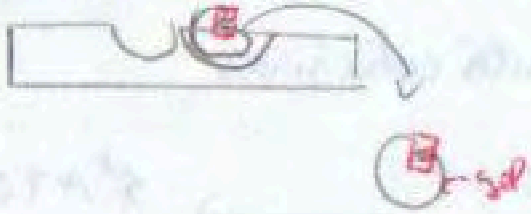
But : séparation de l'ADN en fonction du PM



gel d'Agarose



Bromure d'éthidium : coloration de l'ADN



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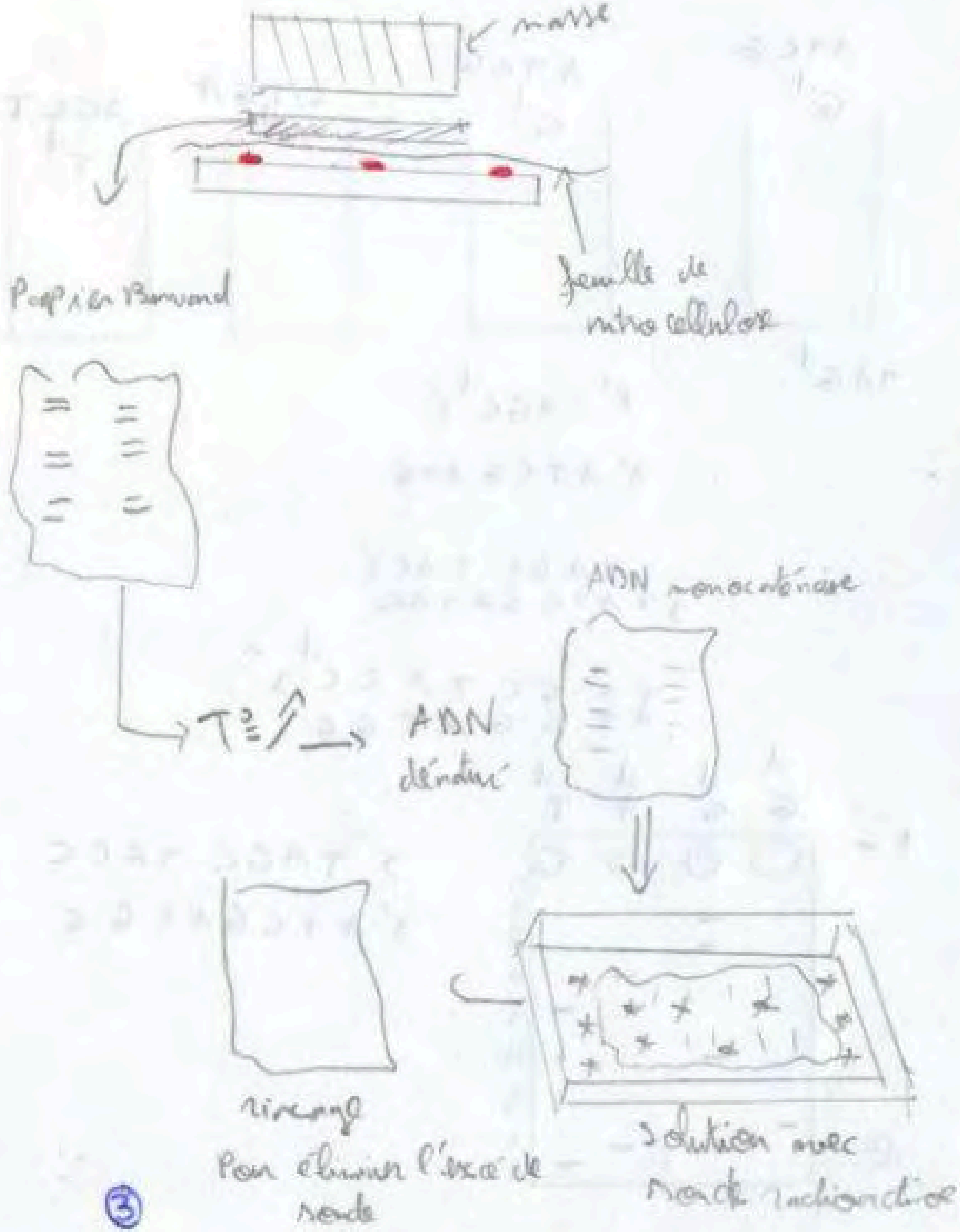
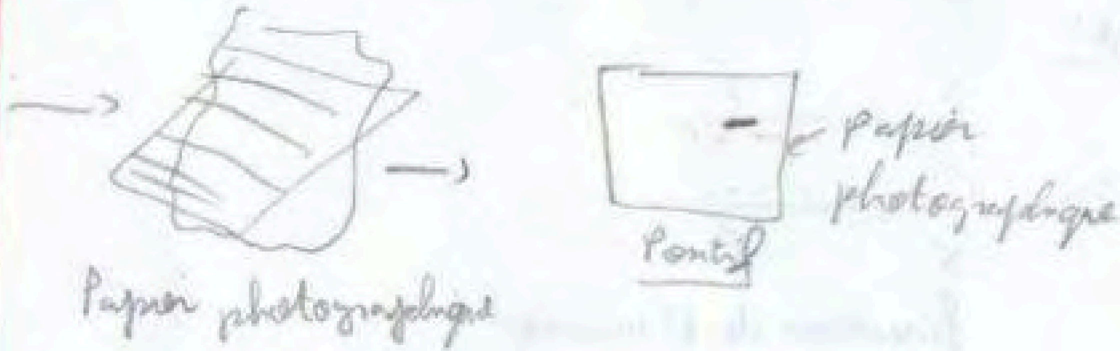
⑦ Thechnique Southern Blot :

Utilisation d'une sonde moléculaire

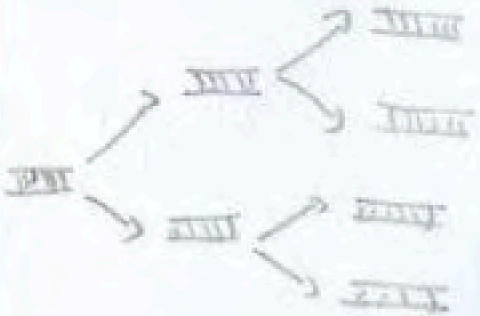




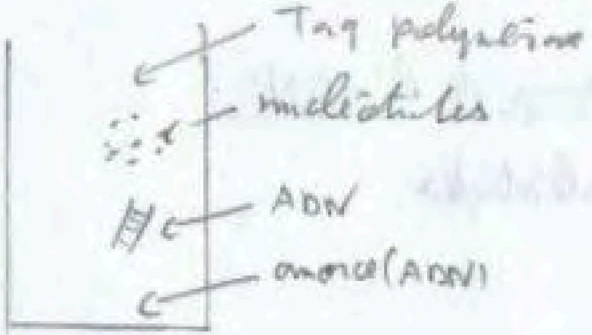
⑧ Thechnique BCR :

Polymérase chain réaction

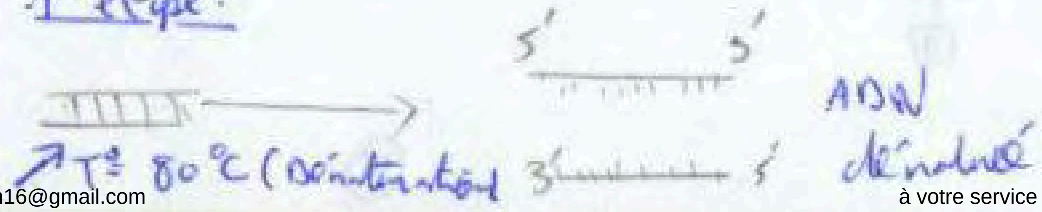
But : amplification de l'ADN



amplification = Ensemble de réplication



1e étape :



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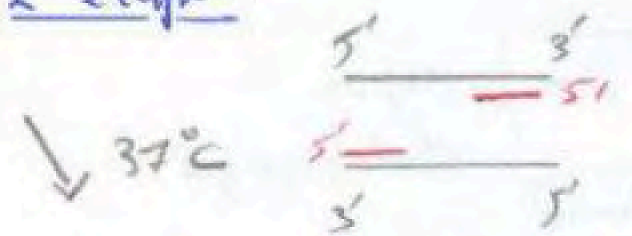
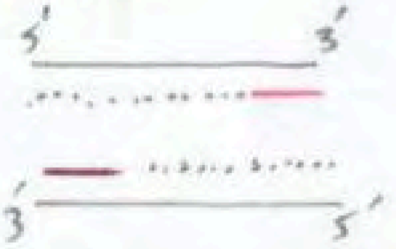
2e étape :



fixation de l'amorce

3e étape :

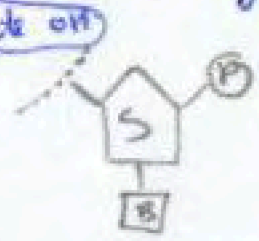


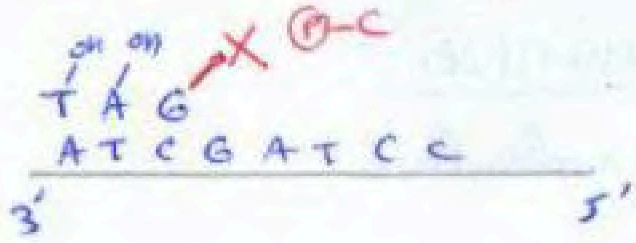
la Taq réplique l'ADN

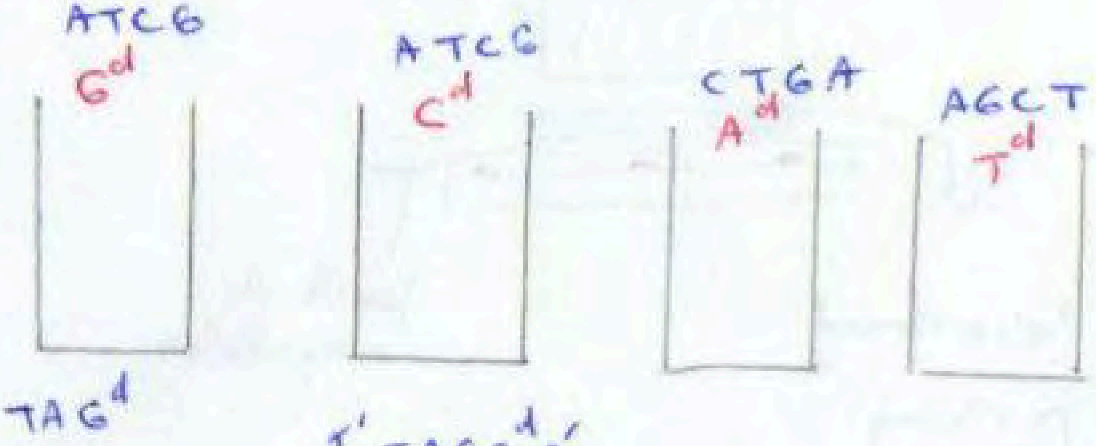
> 3 étapes = 1 cycle
> molécules = 2^n (nbre de cycles)

5) Technique de séquençage de l'ADN :

en 4 de didésoxynucléotides





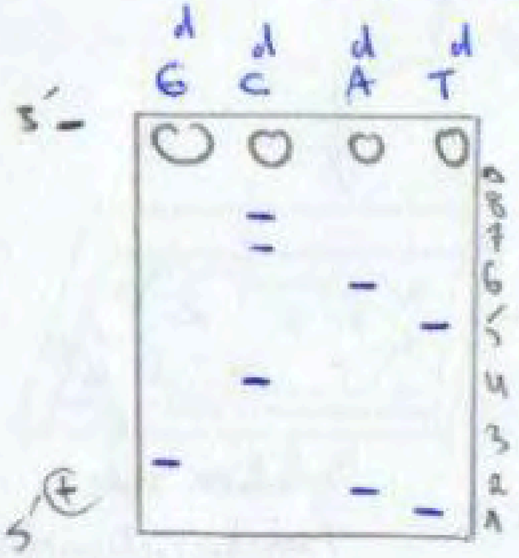


TAG d

5' TAGC d 3'
3' ATCGATG

5' TAGC TAC 3'
3' ATCGATGC

TAGCTACC d 3'
ATCGATGG



5' TAGC TACC
3' ATCGATGG